

Pharmacokinetics Interaction of Glucocorticoids with ^{99m}Tc-MDP Radiopharmaceuticals for Bone Imaging Agents and its Biodistribution Pattern

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Abstract

A drug therapy can alter the pharmacokinetic profiles and biodistribution patterns of radiopharmaceuticals. Glucocorticoids are pharmaceutical drugs for anti-inflammatory by preventing phospholipid release and decreasing eosinophil action. To achieve an optimum diagnostic outcome, this research was focused on pharmacokinetics interaction and biodistribution pattern between two kinds of Glucocorticoids drugs i.e. dexamethasone and prednisone with ^{99m}Tc-MDP using animal model *Mus musculus stock Swiss*. ^{99m}Tc-MDP has been developed as radiopharmaceutical for bone imaging in nuclear medicine. Mice were divided into three groups, which were treated with dexamethasone by oral administration for 5 days continuously, treatment with prednisone by oral administration for 3 days continuously and without treatment (control). Pharmacokinetics interaction was conducted by injecting 200 μ L ^{99m}Tc-MDP intravenously administrated using a dose 1 μ Ci/ μ L. Biodistribution pattern was conducted by injecting 200 μ L ^{99m}Tc-MDP intravenously administrated using a dose 1 μ Ci/ μ L. After 3 hours after intravenous injection of ^{99m}Tc-MDP each of these groups of animals were killed with chloroform and then dissected. Radioactivity of blood samples and selected organs were weighed and counted by using single channel analyzer. The results of pharmacokinetics study showed that the elimination half-life of animal model that given with dexamethasone and prednisone are 4.61 h and 4.63 h more faster than control animals (20.67 h). The results of biodistribution study showed that uptake of ^{99m}Tc-MDP in bone using animal models decreased which were given dexamethasone and prednisone compared to normal animals, which following results $3.53 \pm 0.49\%$, $3.47 \pm 0.5\%$ and $11.54 \pm 4.36\%$ (control).

Key words: ^{99m}Tc-MDP Radiopharmaceuticals, Pharmacokinetics Interaction, Glucocorticoids, biodistribution pattern.

Introduction

^{99m}Tc-MDP radiopharmaceuticals have been clinically approved and widely used for bone imaging in nuclear medicine (Schwochau, 2000). This complexes are preferable because there are more stable in vivo than P-O-P bond of phosphates. P-O-P bond can be easily broken down by phosphatase enzymes, but the P-C-P bond in diphosphonate of ^{99m}Tc-MDP is not affected (Khalil, 2011). This complexes were used for metastatic bone cancer because their high sensitivity can detect metastases before occurrence of anatomical changes (Ogawa & Ishizaki, 2015) (Goyal & Antonarakis, 2012).

Radiopharmaceuticals are used for two purposes. The first is their use as a traced compound administered to a patient for observing physiological alterations or abnormal distribution throughout of the body and the second is their use as a tracer for biochemical or physiological studies (Santos-Oliveira, 2008). The pharmacokinetics or biodistribution of radiopharmaceutical may alter by a variety of drugs, disease states and surgical procedurs (Santos-Oliveira, 2008), which a significant clinical impact on safety, scan interpretation, and diagnostic imaging accuracy (Factors, 2010).

In a previous study, it was known that aluminium containing drugs, nifedipine, etidronate, pamidronate and vitamin D could reduce bone uptake of ^{99m}Tc-MDP and ^{99m}Tc-HDP (Factors, 2010). Glucocorticoids have become one of the most widely used and effective treatments for various inflammatory for palliative treatment. Administration of Glucocorticoids especially for long term systemic by oral or parenteral can induce bone loss, osteoporosis and fractures, adrenal suppression, hyperglycemia, diabetes, cardiovascular disease, and immunosuppression (Liu et al., 2013). This research is focused on studying the Glucocorticoids drug interactions with ^{99m}Tc-MDP radiopharmaceutical using mice stock swiss through observation the changes in pharmacokinetics profiles and biodistribution pattern.

Materials and Methods

The materials to carry out in this research were MDP (Sigma Aldrich), $\text{SnCl}_2 \cdot 2\text{H}_2\text{O}$ (Sigma Aldrich), NaOH (E. Merck), HCl (E. Merck), aquadest (IPHA laboratories), radionuclide Technetium-99m from generator $^{99}\text{Mo}/^{99\text{m}}\text{Tc}$ (POLATOM), Whatman 3MM paper (Whatmann) and pH Universal Indicator (E. Merck). The equipments used were single channel analyzer NaI(TL) detector (Ortec), TLC scanner (Bioscan 2000), Deluxe Isotope Calibration (Victoreen), vial 10 mL, micro pipette (eppendorf), analytical scale (Mettler Toledo), oven (Mettler), disposable syringe (Terumo) and other glass tools.

Animals

This research used 24 male mice (*Mus musculus*) stock Swiss, each weighing 36-44 gram. This research was approved by the ethics Committee for Care and Use of Laboratory Animals (KEPPHP BATAN) with ethical approval number 003/KEPPHP-BATAN/IV/2015 and was performed according to guidelines on animal experimentation.

Statistical Analysis

The data were calculated to an unpaired, two-tailed distribution student t-test by statplus program, with 95% of confidence level.

Radiolabeling of $^{99\text{m}}\text{Tc}$ -MDP and determination of radiochemical purity (Zolle, 2007)

The freeze-dried MDP kit was reconstituted using 1 mL of freshly eluted $^{99\text{m}}\text{TcO}_4$ solution containing 1-3 mCi of radioactivity. The solution was stirred for 1 minute and incubated 30 minutes at the room temperature. The radiochemical purity was determined by ascending paper chromatography method using acetone and saline as mobile phase and whatmann 3MM as stationary phase. Paper chromatogram was dried in oven at 80°C for 5 minutes and then every 1 cm piece of paper is measured by the single-channel gamma counter detector NaI (TI).

Pharmacokinetic Study

The pharmacokinetic study were performed by injecting 0.1 mL of $^{99\text{m}}\text{Tc}$ -MDP intravenously with a dose 1 $\mu\text{Ci}/\mu\text{L}$ in animals group I, II and III, which each group consists of three animals. The blood samples were collected from animal tail at 0,833; 1; 2; 3; 4; 5; 6; 24; 25; 26 hours after injection of $^{99\text{m}}\text{Tc}$ -MDP. The blood samples were weighed and then the radioactivity was measured using a single channel analyzer with Na(I)TL detector. Samples were calculated and analyzed to get percentage of injection dose per gram samples (%ID/g) and the half life ($t_{1/2}$) of distribution and elimination of $^{99\text{m}}\text{Tc}$ -MDP were calculated using Multifit software (version 8/2000, developed by Dr. J.H.Proost, University Center for Pharmacy, Groningen, The Netherlands).

Biodistribution study

Normal animal group I and animal models group II and III were injected with 0.1 mL of $^{99\text{m}}\text{Tc}$ -MDP with a dose of 1 $\mu\text{Ci}/\mu\text{L}$ intravenously through the tail. Then, 3 hours after intravenous injection, the animals were killed with chloroform and the vital organs were taken such as muscle, bone, blood, intestine, stomach, liver, spleen, kidneys, heart and lungs. Samples were collected, weighed, and the radioactivity of each samples were counted using single channel analyzer with Na(I)TL detector. The accumulation of each organ samples were calculated as percentage of injection dose per gram organ.

Results and Discussion

Radiochemical purity is one of the requirements of radiopharmaceutical quality testing in addition to clarity, vacuum and pH. Radiochemical purity was determined by ascending paper chromatography system. As shown in Figure 1, acetone as a mobile phase was used to separate pertechnetate and saline (NaCl 0.9%) was used to separate the reduced technetium, then the percentage of radiochemical purity was calculated based on a calculation formula of $100\% - \%(\text{TcO}_4^- + \text{TcO}_2)$. The results of this test showed that $^{99\text{m}}\text{Tc}$ -MDP radiopharmaceuticals had $97.92 \pm 1.93\%$ radiochemical purity ($n=5$) with impurity of TcO_2 was $1.88 \pm 1.92\%$ and TcO_4^- was $0.71 \pm 1.18\%$. The $^{99\text{m}}\text{Tc}$ -MDP radiopharmaceuticals have been fulfilled the requirement of United State of Pharmacopoeia, which the percentage of radiochemical purity is greater than 95%. The solution of MDP dissolved easily in saline, giving a clear and colorless solution, vacuum and pH between 5.8 – 6.0.

The pharmacokinetic profile was conducted to determine the rate of distribution and elimination of $^{99\text{m}}\text{Tc}$ -MDP radiopharmaceutical in the body through the determination of its biological half life. As shown in Figure 2, percentage of injection dose of $^{99\text{m}}\text{Tc}$ -MDP in animal models treatment dexamethasone and prednisone for several days showed an increase compared to the control, results at $13.28 \pm 2.79\%$ ID/g and $13.56 \pm 3.85\%$ ID/g, respectively than control at $2.08 \pm 0.36\%$ ID/g. Treatment of dexamethasone and prednisone was similar to pharmacokinetic profile (Fig. 2).

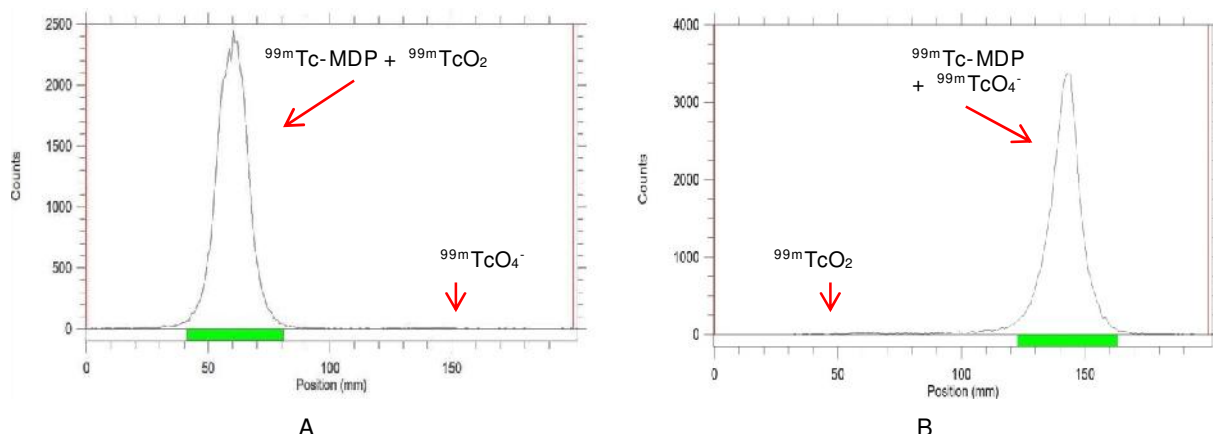


Figure 1. The chromatogram of ^{99m}Tc -MDP using (A) acetone as mobile phase and (B) saline as mobile phase

About 12.34 and 12.06% of the injected dose remains in the blood at 1 hour post-injection and 0.19 and 0.18% at 24 hours, ^{99m}Tc -MDP were treatment with dexamethasone and prednisone. Following intravenous administration, ^{99m}Tc -MDP is cleared from the plasma with a half-time of 3–4 min. About 10% of the injected dosage remains in the blood at 1 hour post-injection and less than 1% at 24 hours (Saha, 2010).

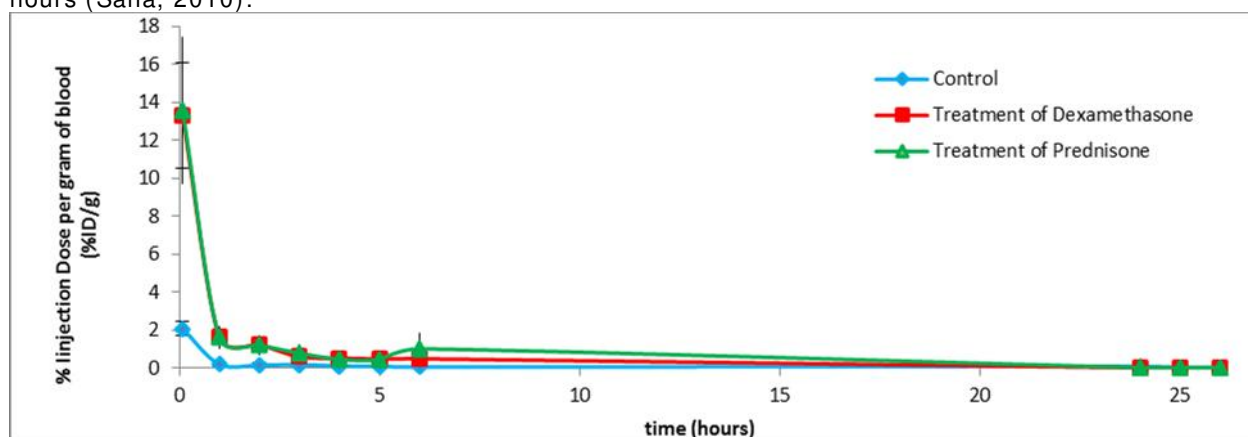


Figure 2. Pharmacokinetics profile of ^{99m}Tc -MDP using normal mice stock swiss with treatment of dexamethasone and prednisone and without treatment as control.

Based on calculations by the multifit software (Table 1), it showed that the distribution half-life of ^{99m}Tc -MDP with treatment did not differ significantly from control. But the elimination half-life showed a significant difference where the biological half-life of ^{99m}Tc -MDP radiopharmaceutical which treatment with dexametasone (4.61 h) and prednisone (4.43 h), the half-life more faster than control (20.67 h).

Table 1. Distribution and elimination half-life of ^{99m}Tc -MDP radiopharmaceutical given Glucocorticoids drug treatment compared to controls without treatment using Multifit software

	$t_{1/2}$ distribution (hours)	$t_{1/2}$ elimination (minutes)
Controls (without treatment)	0.21	20.67
Treatment of Dexamethasone	0.23	4.61
Treatment of Prednisone	0.20	4.43

Biodistribution study was performed 3 hours after intravenous injection of ^{99m}Tc -MDP to animal model. In nuclear medicine, imaging of bone scintigraphy using ^{99m}Tc -MDP usually performed 2-5 hours afterwards for clearance of the administrated radiopharmaceutical from the intravascular compartment and from the extracellular nonosseous soft tissues (Zuckier & Martineau, 2015).

The administration of dexamethasone and prednisone for several days has changed the pattern of biodistribution particularly on the bone. As shown in Figure 3, the highest percentage shown in the bone, but the bones which given treatment with dexamethasone and prednisone reduced bone uptake than control, results were 3.53 ± 0.49 % and 3.47 ± 0.5 %, respectively which compared to the control 11.54 ± 4.36 % (t test, differ significantly with the 95% confidence level). Dexamethasone and prednisone, glucocorticoids agent, are common to control and treat several inflammatory diseases and side effect of this agents especially for long term is bone loss through decrease quality and quantity of mineral bone density and rate of osteogenesis (Doroudi et al., 2012). A small amount of bone synthesis depends on decreased calcium availability and decreased glucocorticoid-induced osteoblastic activity (Longui, 2007). Based on t-test, uptake of non target organs showed there are no significantly differences between treatments and controls. Non target organs have lower uptake than bone (target organ), which are below than 2.5% of injected dose per gram.

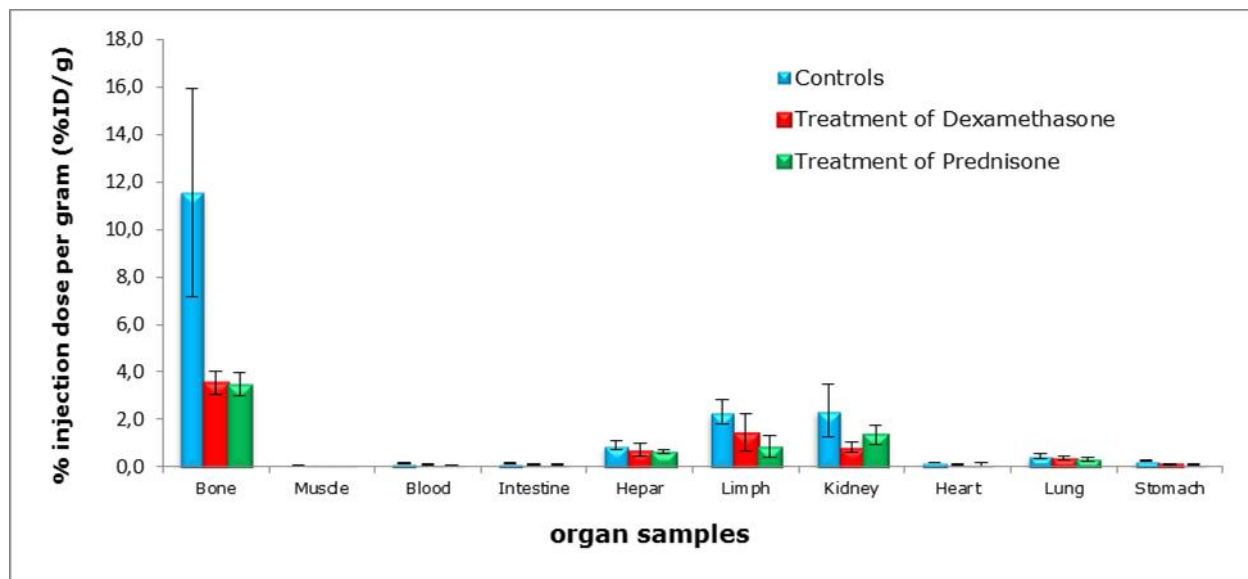


Figure 3. Biodistribution profile of ^{99m}Tc-MDP using normal mice stock swiss with treatment of dexamethasone and prednisone and without treatment

Conclusions

Administration of dexamethasone and prednisone as glucocorticoid agent can alter pharmacokinetics profile and biodistribution pattern. Treatment of dexamethasone and prednisone decreases bone uptake and elimination half-life of ^{99m}Tc-MDP radiopharmaceuticals

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